# Some Aquatic Hyphomycetes of Florida

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THE aquatic Hyphomycetes are imperfect fungi found in the form-family Moniliaceae because of their hyaline conidia and conidiophores. These organisms occur in freshwater streams that are typically fast moving and well oxygenated. They have also been found in polluted streams (Conway, 1968). Aquatic Hyphomycetes are usually found in association with well decayed leaves, although they have been found on other substrate (Nilsson, 1964).

The aquatic Hyphomycetes were first described in 1893 by De-Wildeman. However, active research concerning these organisms did not begin until Ingold (1942) published his first of many papers on the fungal flora of England.

Since Ingold's first report most of the world's land masses have been explored: Africa (Dixon, 1959; Ingold, 1956), Asia (Tubaki, 1957, 1965), Australia (Cowling and Waid, 1963), Canada (Ingold, 1960), Europe (Banhegyi, 1962; Ingold, 1949; Fenton, 1950; Nilsson, 1964), South America (Nilsson, 1962), and the United States of America (Baxter, 1961, 1964; Conway, 1968; Crane, 1968; Petersen, 1960, 1962, 1963a, 1963b; Ranzoni, 1953; and Umphlett, 1959).

Published reports from the United States have been concerned principally with the northeastern and western United States. This is the first report of aquatic Hyphomycetes from Florida.

### TAXONOMIC CRITERIA

Classification of aquatic Hyphomycetes is based primarily on conidial development and secondarily on conidial form.

The two main types of development are the aleuriosporeae and the phialosporae. These are defined as:

- 1) Aleuriosporae: A conidium is delimited from its conidiophore early in its development. It is termed an aleuriospore or terminal chlamydospore. The aleuriospore arises as a modified terminal portion of a hypha.
- 2) Phialosporae: A conidium borne on a phialide is termed a phialospore. A phialide is a unicellular flask shaped struc-

ture with a comparatively enlarged venter and a neck-like portion which bears the conidium at or within the tip. The phialospore is not delimited from the phialide until maturity.

Essentially three forms exist in the aquatic Hyphomycetes: ovoid, sigmoid and tetraradiate. It was thought at first that these forms represented evolution of increasing efficiency of floatation and entanglement. However, Webster (1959a) has demonstrated that all settle at approximately the same rate, but that the tetraradiate form is more efficient in entanglement.

Although these organisms are Fungi imperfecti, five aquatic Hyphomycetes have been found to possess a sexual stage (Ranzoni, 1956; Tubaki, 1966; Webster, 1959b, 1961, 1965). Anguillospora crassa Ingold represents the conidial stage of a species of Mollisia, Flagellospora penicillioides Ingold and Heliscus lugdunensis Saccardo and Therry represent conidial stages of Nectria, Dactylella aquatica represents a conidial stage of Massarina aquatica Webster, and Varicosporium sp. represents a conidial stage of Hymenoscyphus.

### MATERIAL AND METHODS

Aquatic Hyphomycetes are best observed from well decayed leaves. These leaves can be collected from the streams along with a water and foam sample. The foam acts as a natural trap for conidia and gives a fairly accurate indication of the fungal flora of the stream.

The samples are brought back to the laboratory and observed with the 10X lens of a microscope. The sample is placed in an open petri plate and conidia can be seen either floating on the surface of the water or on the bottom of the plate. If cultures are desired, conidia can be picked up using a small diameter pasteur pipette. The conidia are then plated on 1 per cent water agar and allowed to germinate for 24 hours. These must then be retransferred or they will become over-grown by bacteria and other fungi.

The cultures will not produce typical conidia until they are re-immersed in water. Therefore, for verification of the isolation sections of the colony are put back into water contained in a sterile petri plate. If the isolation was successful, typical aquatic conidia will develop in three to five days. Material for this investigation has been taken principally from the streams occurring at the Devil's Millhopper which is located approximately six miles northwest of Gainesville, Florida.

# Results

The aquatic Hyphomycetes are represented by a world flora of 70 species and 38 genera. In this investigation 15 species from 12 genera and one undescribed conidial type have been found. Listed below are the aquatic Hyphomycetes found from Florida.

1. Actinospora megalospora Ingold, 1952. Conidia are tetraradiate aleuriospores (Fig. 1A). Measurements are: center cell 35- $50\mu$  in diameter, 4-8 lateral arms  $100-180\mu \times 8\mu$ . The center cell is usually of a foamy, brown appearance and the lateral arms are hyaline. These are the largest conidia found in this group. This species was obtained commonly from the streams. Distribution in the USA: Conn. (Crane), Florida (Conway), New York (Conway).

2. Anguillospora gigantea Ranzoni, 1953. Conidia are the largest in the genus Anguillospora (Fig. 1C). They are multiseptate sigmoid aleuriospores which measure  $350-700\mu \times 5-7\mu$ . The species has only been found in great abundance in April 1969. Distribution in the USA: Calif. (Ranzoni), Florida (Conway), New York (Petersen).

3. Anguillospora longissima (Sacc. & Syd.) Ingold, 1942. This is a multiseptate sigmoid aleuriospore measuring  $150-300\mu \times 5-7\mu$ (Fig. 1D). It has been found to be common in Florida. The mechanism of release thought to occur only in this species is characterized by the collapse of a separating cell beneath the conidium, which releases the conidium. Distribution in the USA: Calif. (Ranzoni), Conn. (Petersen), Florida (Conway), Ga. (Petersen), Ind. (Baxter), Mass. (Ingold), N. J. (Peterson), N.Y. (Conway, Petersen), Pa. (Petersen), S. C., Tenn. (Petersen), Va. (Umphlett), W. Va. (Crane), Wyoming (Baxter).

4. Anguillospora pseudolongissima Ranzoni, 1953. This species is very similar to A. longissima except that conidia never exceed  $100\mu$  in length (Fig. 1B). Its occurrence in Florida is rare. Distribution in the USA:Calif. (Ranzoni), Conn. (Petersen), Fla. (Conway), Ga. (Petersen), N.Y. (Conway), Pa., S.C., Tenn. (Petersen).



Fig. 1. A, Actinospora megalospora  $\times$  100; B, Anguillospora pseudolongissima  $\times$  500; C, Anguillospora gigantea  $\times$  500; D, Anguillospora longissima  $\times$  500.

5. Articulospora tetracladia Ingold forma tetracladia Nilsson, 1954. The aleuriospore consists of four arms (Fig. 2D). The first arm is 16-25  $\mu \times 3$ -5  $\mu$  and consists of 2-3 cells. This arm is usually characterized by the presence of a slight inflation near the point of divergence of the arms. The other three arms are approximately equal in length 20-50  $\mu \times 3$ -4  $\mu$ . Conidia of this species have been found in only a few collections. Distribution in the USA: Calif. (Ranzoni), Conn. (Petersen), Del. (Crane), Fla. (Conway), Ga. (Petersen), Mass. (Nilsson), Md., Me. (Crane), N.C. (Petersen), N.H. (Crane, Nilsson), N.J. (Petersen), N.Y. (Conway, Petersen), Pa., S.C., Tenn. (Petersen), Va., Vt., W. Va. (Crane), Wyo. (Baxter).

6. Campylospora chaetocladia Ranzoni, 1953. This is an unusual crozier form of a multiseptate tetraradiate arrangement (Fig. 2B). It is an aleuriospore and is commonly found in the streams of this area. Overall dimensions of a conidium from arm to arm is approximately 100  $\mu$ . Distribution in the USA: Calif. (Ranzoni), Fla. (Conway), N.Y. (Petersen), Oreg. (Baxter), W. Va. (Crane).

7. Clavatospora tentacula (Umphlett) Nilsson, 1964. Conidia are unicellular, tetraradiate phialospores. The main axis is clavate 56-62  $\mu$  in length (Fig. 2C). Slender lateral arms 30-55 $\mu$  in length arise near the apex of the clavate head. It is common in Florida. Distribution in the USA: Fla. (Conway), Ind. (Baxter), Va. (Umphlett), N.Y. (Conway).

8. Flagellospora curvula Ingold, 1942. Conidia are unicellular sigmoid phialospores measuring up to 180  $\mu$  in length (Fig. 2E). This species is characterized by having long slender conidia and from two to ten phialides per conidiophore. This species has been observed only a few times from this area. Distribution in the USA: Calif. (Ranzoni), Fla. (Conway), Mass. (Nilsson), N.C. (Petersen), N.H. (Nilsson), N.Y. (Conway).

9. Geniculospora inflata (Ingold) Nilsson, 1964. Conidia are tetraradiate aleuriospores (Fig. 2A). Each arm measures 40-120  $\mu \times 3$ -4  $\mu$  and is usually 2-3 septate. Conidia are usually characterized by an inflation at the point of divergence of the arms. Its occurrence in Florida is rare. Distribution in the USA: Fla. (Conway), Me. (Crane), N.J. (Petersen), N.Y. (Conway, Petersen), S.C. (Petersen).

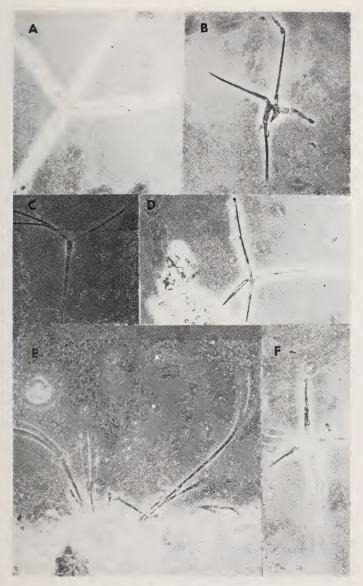


Fig. 2. A, Geniculospora inflata; B, Campylospora chaetocladia; C, Clavatospora tentacula; D, Articulospora tetracladia forma tetracladia; E, Flagellospora curvula; F, Lemonniera aquatica. All ×500.

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10. Lemonniera aquatica DeWild., 1894. This species is the largest tetraradiate phialospore (Fig. 2F). The four arms of a conidia are 20-70  $\mu \times 3$ -4  $\mu$ . Conidia are inserted on the phialide not at the tip of one of the arms but at the point of divergence of the arms. This species was common in occurrence. Distribution in the USA: Calif. (Ranzoni), Fla. (Conway), Mass. (Nilsson), N.C. (Petersen), N.H. (Nilsson), N.J. (Petersen), N.Y. (Conway, Petersen), Pa., S.C., Tenn. (Petersen), Va. (Crane, Umphlett), W. Va. (Crane), Wyo. (Baxter).

11. Lunulospora curvula Ingold, 1942. Conidia are unicellular crescent shaped aleuriospores measuring 66-90  $\mu \times 5$ -6  $\mu$  (Fig. 3A). Conidia may appear septate due to the presence of vacuoles. Point of attachment of the conidia is not at the tip, but toward the middle of the convex side. The conidia are shed by the breakdown of a separating cell. This is one of the more common species in this area. Distribution in the USA: Calif. (Ranzoni), Conn. (Petersen), Del. (Crane), Fla. (Conway), Ga. (Petersen), Mass. (Nilsson), N.C. (Petersen), N.H. (Nilsson), N.Y. (Conway, Petersen), Oreg. (Baxter), Pa., S.C. (Petersen), Va. (Umphlett), Wyo. (Baxter).

12. Tetrachaetum elegans Ingold, 1942. This is a large multiseptate tetraradiate aleuriospore (Fig. 3E). Its main axis may exceed 200  $\mu$ , with each arm measuring 120-150  $\mu \times 2$ -4  $\mu$ . Point of attachment to the conidiophore is at the tip of one of the arms. It is common in occurrence in this area. Distribution in the USA: Calif. (Ranzoni), Conn. (Petersen), Fla. (Conway), N.C. (Petersen), N.H. (Nilsson), N.J. (Petersen), N.Y. (Conway, Petersen), Oreg. (Baxter,) Pa., S.C., Tenn. (Petersen), Va. (Crane, Umphlett), Wyo. (Baxter).

13. Tetracladium marchalianum DeWild., 1893. Conidia are tetraradiate aleuriospores (Fig. 3C). Three arms are 24-30  $\mu \times 2$ -3  $\mu$  and the basal extension 16-27 $\mu \times 4$ -5  $\mu$ . Conidia are characterized by the presence of two protuberances at the divergence of the arms. The primary protuberance is 5-7  $\mu \times 8$ -9  $\mu$  and the secondary protuberance is approximately 5  $\mu$  in diameter. This is the most frequently occurring aquatic Hyphomycete in Florida. Distribution in the USA: Calif. (Ranzoni), Del. (Crane), Fla. (Conway), Mass. (Nilsson), N.C. (Petersen), N.H. (Nilsson), N.J.

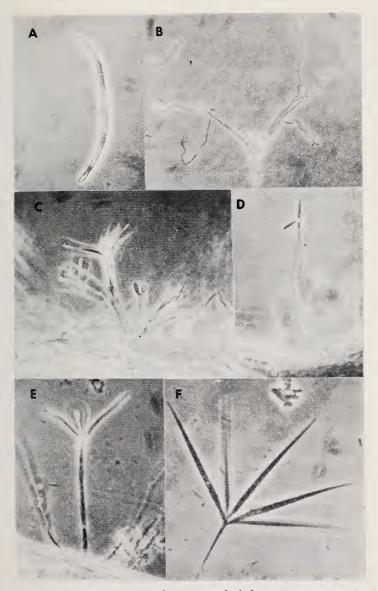


Fig. 3. A, Lunulospora curvula; B, Tetraclacladium setigerum, germinating conidia; C, Tetracladium marchalianum; D, Tricelophorus monosporus; E, Tetrachaetum elegans; F, Unknown conidia. All  $\times 500$ .

(Petersen), N.Y. (Conway, Petersen), Oreg. (Baxter), Va. (Umphlett), W. Va. (Crane), Wyo. (Baxter).

14. Tetracladium setigerum (Grove) Ingold, 1942. The conidia are similar to *T. marchalianum* except for the presence of three protuberances at the divergence of the arms (Fig. 3B). Each protuberance is approximately 11-15  $\mu \times 4$ -5  $\mu$  and may be 0-3 septate. This species occurred rarely and was only present in one collection in April 1969. Distribution in the USA: Calif. (Ranzoni), Fla. (Conway), N.C. (Petersen), N.Y. (Conway, Petersen), S.C. (Petersen), Va. (Umphlett).

15. Triscelophorus monosporus Ingold, 1943. Conidia are solitary aleuriospores with the main axis of the conidia being an extension of the short conidiophore (Fig. 3D). The main axis is 30-50  $\mu \times 4$ -5  $\mu$  at its widest part, and the lateral branches are 16-30  $\mu \times 2.0$ -2.5  $\mu$ . The three lateral arms arise one at a time as buds from the main axis. This species is common in this area. Distribution in the USA: Calif. (Ranzoni), Fla. (Conway), Ind. (Baxter), N.Y. (Conway, Petersen), Tenn. (Petersen), Va. (Umphlett).

16. Undescribed conidial type. The unknown conidium consists of a main axis 38  $\mu$  in length which tapers in width to 3-0.5  $\mu$  from the point of attachment of the arms to the conidiophore (Fig. 3F). The arms are up to 85  $\mu$  in length. They are constricted at their attachment to the main axis, but enlarge to 4  $\mu$  in width at approximately one-third of their length and then taper to 0.5-1  $\mu$  at their tips. The main axis is two septate. The arms are 11-12 septate and appear to be constricted only at septa in the enlarged portion of the arms. Obtaining this organism in pure culture is necessary for further description.

### Conclusion

Although the area sampled is small in relation to the entire state, this technique of continuous sampling has been shown to be a satisfactory, if not superior, method of determining presence of species for a larger area (Conway 1968). The reason for this appears to be that aquatic Hyphomycetes may be cyclic in occurrence, being present in the streams only under certain conditions. Therefore, one sampling of an area would be a poor criterion of species present in that area.

#### CONWAY: Aquatic Hyphomycetes of Florida

As a result of collections over a nine month period, certain tendencies of the aquatic flora can be observed. *Tetracladium marchalianum* and *Actinospora megalospora* are the most frequently occurring species in this area. *Lunulospora curvula*, *Anguillospora longissima* and *Triscelophorus monosporus* are also very common. Species such as *Anguillospora gigantea* and *Tetracladium setigerum* occur sporadically, but when present they are in great numbers.

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